

What Is Claimed Is:

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1. A flow-through device for capturing a target nucleic acid comprising a three-dimensional porous substrate having immobilized thereon a capture polynucleotide which is capable of hybridizing to the target nucleic acid.

2. The flow-through device of Claim 1, which is about 1 mm to 20 mm thick.

3. The flow-through device of Claim 1, in which said porous substrate has an average pore size of about 1 μm to about 250 μm .

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4. The flow-through device of Claim 1, in which said porous substrate has immobilized thereon about 2×10^{-19} to 2×10^{-15} nmole/nm² of said capture polynucleotide.

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5. The flow-through device of Claim 1, in which said capture polynucleotide is covalently attached to the porous substrate.

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6. The flow-through device of Claim 1, in which said capture polynucleotide is covalently attached to the porous substrate via a phosphodiester, phosphorothioate or phosphoramidate linkage.

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7. The flow-through device of Claim 1, in which said capture polynucleotide is covalently attached to the porous substrate via a carboxamide linkage.

8. The flow-through device of Claim 1, in which said capture polynucleotide is covalently attached to the porous substrate via a linker.

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9. The flow-through device of Claim 1, in which said porous substrate is composed of glass or a polymeric material selected from the group consisting of polyethylene, polystyrene, polycarbonate and polypropylene.

10. The flow-through device of Claim 1, in which said porous substrate is composed of high density or high molecular weight polyethylene.

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11. The flow-through device of Claim 1, in which said porous substrate has a void volume in the range of about 1 $\mu\text{l}/\text{cm}^2$ to about 100 $\mu\text{l}/\text{cm}^2$.

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12. A flow-through device for capturing a target nucleic acid comprising a three-dimensional substrate having an average pore size of about 1 μm to about 250 μm and having immobilized thereon a capture polynucleotide capable of hybridizing to the target nucleic acid.

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13. The flow-through device of Claim 12, which has a porosity in the range of about 25 to 80%.

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14. The flow-through device of Claim 12, in which the capture polynucleotide is covalently immobilized on the porous substrate via its 5'- or 3'- terminal residue.

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15. The flow-through device of Claim 14 further including a linker disposed between the porous substrate and the capture polynucleotide.

16. The flow-through device of Claim 12, having immobilized thereon about 2×10^{-19} to 2×10^{-15} nmole/ nm^2 of said capture polynucleotide.

17. A flow-through device for capturing a target nucleic acid comprising a three-dimensional porous substrate having immobilized thereon about 2×10^{-19} to 2×10^{-15} nmole/nm² of a capture polynucleotide capable of hybridizing to the target nucleic acid.

18. The flow-through device of Claim 17 in which the porous substrate has an average pore size of about 1 μ m to about 250 μ m.

19. The flow-through device of Claim 17 in which the porous substrate has a porosity in the range of about 25 to 80%.

20. The flow-through device of Claim 17 in which said capture polynucleotide is covalently attached to the porous substrate.

21. An apparatus for capturing a target nucleic acid from a sample comprising a housing having disposed therein a flow-through device according to Claim 1,

22. The flow-through device of Claim 21, in which said housing is selected from the group consisting of a syringe barrel, a pipette, a disposable pipette tip, a chromatography column, a spin column, a microchannel, a capillary and a multi-well plate.

23. A method of capturing a target nucleic acid from a sample, said method comprising the step of:

(i) flowing a sample containing or suspected of containing a target nucleic acid through a three-dimensional porous substrate having immobilized thereon a capture polynucleotide capable of hybridizing to the target nucleic

acid, under conditions wherein said target nucleic acid and capture polynucleotide hybridize to one another to form a hybridized complex, thereby capturing the target nucleic acid.

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10 24. The method of Claim 23, in which said target nucleic acid is applied to said porous substrate under conditions of high stringency.

25. The method of Claim 23, in which said target nucleic acid is applied to said porous substrate under conditions of low stringency.

26. The method of Claim 23, in which said target nucleic acid is applied to the porous substrate under conditions
15 wherein it hybridizes with said capture polynucleotide in less than one minute.

27. The method of Claim 23, in which said porous substrate has an average pore size of about 1 μm to about 250
20 μm .

28. The method of Claim 23, in which the density or surface concentration of said capture polynucleotide is about
25 2×10^{-19} to 2×10^{-15} nmole/nm².

29. The method of Claim 23, in which said capture polynucleotide is covalently attached to the porous substrate.

30 30. The method of Claim 23, in which said capture polynucleotide is covalently attached to the porous substrate via a phosphodiester, phosphorothioate or phosphoramidate linkage.

31. The method of Claim 23, in which said capture polynucleotide is covalently attached to the porous substrate via a carboxyamide linkage.

32. The method of Claim 23, in which said capture polynucleotide is covalently attached to the porous substrate via a linker.

33. The method of Claim 23, in which said porous substrate is composed of glass or a polymeric material selected from the group consisting of polyethylene, polystyrene, polycarbonate and polypropylene.

34. The method of Claim 23, in which said porous substrate is composed of high density or high molecular weight polyethylene.

35. The method of Claim 23, in which said porous substrate has a void volume in the range of $0.1 \mu\text{l}/\text{cm}^2$ to about $100 \mu\text{l}/\text{cm}^2$.

36. The method of Claim 23 which further includes the step of washing said hybridized complex under conditions of moderate or high stringency.

37. The method of Claim 23 which further includes the step of dissociating the hybridized complex and recovering the target nucleic acid.

38. A method of recovering a target nucleic acid from a sample, said method comprising the steps of:

(a) flowing a sample containing or suspected of containing a target nucleic acid through a three-dimensional porous substrate having attached thereto a capture

polynucleotide capable of hybridizing to the target nucleic acid, under conditions wherein the target nucleic acid and target polynucleotide hybridize to form a hybridized complex; and

5 (b) dissociating the hybridized complex and recovering said target nucleic acid.

10 39. The method of Claim 38, which further includes the step of washing the porous substrate under conditions of high stringency following step (a) and prior to step (b).

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15 40. A method of determining whether a sample contains a target nucleic acid, said method comprising the steps of:
(c) flowing a sample suspected of containing a target nucleic acid through a three-dimensional porous substrate having a capture polynucleotide capable of hybridizing to the target nucleic acid attached thereto under conditions wherein the target nucleic acid and target polynucleotide hybridize; and

20 (d) detecting the presence of hybrids, wherein a positive detection indicates the presence of the target nucleic acid in the sample.

25 41. The method of Claim 40, in which said target nucleic acid bears a reporter moiety and hybrids are detected by detecting the presence of said reporter moiety.

30 42. A method of capturing a sequencing ladder of polynucleotides generated from a sequencing reaction comprising flowing a sample containing sequencing ladder polynucleotides through a porous substrate having immobilized thereon a capture polynucleotide capable of hybridizing to the sequencing ladder polynucleotides under conditions wherein

said capture polynucleotide and said sequencing ladder polynucleotides hybridize.

43. The method of Claim 42, further including the step of washing said porous substrate after said hybridization step.

44. A kit for capturing a target nucleic acid of interest from a sample, comprising:

- a) a three-dimensional porous substrate having immobilized thereon a capture polynucleotide capable of hybridizing to said target nucleic acid; and
- b) a housing into which the porous substrate can be disposed.

45. A kit for sequencing a target nucleic acid of interest comprising:

- a) a forward sequencing primer capable of hybridizing to the target nucleic acid; and
- b) a three-dimensional porous substrate having immobilized thereon a capture polynucleotide capable of hybridizing to a first plurality of sequencing ladder polynucleotides.

46. The kit of Claim 45, further including means for generating a sequencing ladder of polynucleotides from the target nucleic acid.

47. The kit of Claim 45 further including a reverse sequencing primer capable of hybridizing to the target nucleic acid and a second three-dimensional porous substrate having immobilized thereon a second capture polynucleotide capable of hybridizing to a second plurality of sequencing ladder polynucleotides.

48. The kit of Claim 47, in which the forward and reverse sequencing primers have the formula: A-linker-B wherein:

A is a first polynucleotide sequence capable of hybridizing to the capture polynucleotide;

B is a second polynucleotide sequence capable of hybridizing to the target nucleic acid;

"linker" is polyethylene glycol containing from 1 to 10 ethylene glycol units; and

each "-" independently represents a phosphodiester, phosphorothioate or amide linkage.

49. The kit of Claim 48, in which the forward and reverse sequencing primers are as depicted in Fig. 1.

50. A kit for capturing a target nucleic acid from a sample comprising:

a) a three-dimensional porous substrate activated with about 6×10^{-17} to 9×10^{-15} nmol/nm² of a reactive group; and

b) a capture polynucleotide capable of being covalently attached to the porous substrate.

51. The kit of Claim 50, further including a linker capable of being covalently attached to the porous substrate and the capture polynucleotide.

52. A kit for capturing a target nucleic acid from a sample comprising:

a) a porous substrate capable of being activated with about 6×10^{-17} to 9×10^{-15} nmol/nm² reactive groups; and

b) means for generating a capture polynucleotide which is capable of hybridizing to the target nucleic acid and which is capable of being covalently attached to the porous substrate.

53. An apparatus for capturing a target nucleic acid from a sample comprising a housing having disposed therein a flow-through device according to Claim 12.

5 54. The flow-through device of Claim 53, in which said housing is selected from the group consisting of a syringe barrel, a pipette, a disposable pipette tip, a chromatography column, a spin column a microchannel, a capillary and a multi-well plate.

10 55. An apparatus for capturing a target nucleic acid from a sample comprising a housing having disposed therein a flow-through device according to Claim 17;

15 56. The flow-through device of Claim 55, in which said housing is selected from the group consisting of a syringe barrel, a pipette, a disposable pipette tip, a chromatography column, a spin column a microchannel, a capillary and a multi-well plate.

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